Application Serial No. 60/079,769 entitled "Nanomolar, Non-Peptide Inhibitors of Plasmepsin," filed on March 24, 1998 ~~and bearing Attorney Docket No. 02307Z-0085320~~, the teachings of which are incorporated herein by reference.

II. EXAMPLE II

A. Assays

1. Preparation and Maintenance of Entorhinohippocampal Slice Cultures

Organotypic entorhinohippocampal cultures were prepared using the technique of Stoppini, *et al.*, *J. Neurosci. Methods*, **37**, 173-182 (1991). Briefly, the caudal pole of the cerebral hemisphere containing the entorhinal cortex and hippocampus were harvested from brains of 6-7 days old Sprague-Dawley rat pups under sterile condition. 400 μm horizontal entorhinohippocampal sections cut vertical to the long axes of hippocampus were obtained using a McIlwain tissue chopper in a cutting medium consisting of MEM (with Earle's salts, Gibco), 25 mM HEPES, 10 mM Tris Base, 10 mM Glucose, and 3 mM $MgCl_2$ (pH 7.2). Brain tissue explants were then planted onto 30 mm cell culture inserts (Illicell-CM, Millipore, Bedford, MA) that were placed in 6 well culture trays with 1 mL of growth medium (MEM with Hank's salts, Gibco, 20% horse serum, 3 mM glutamine, 25 mM HEPES, 5 mM $NaHCO_3$, 25 mM glucose, 0.5 mM ascorbate, 2 mM $CaCl_2$, 2.5 mM $MgCl_2$, 0.5 mg/L insulin, and penicillin, pH 7.2; Bi, *et al.*, *J. Comp. Neuro.*, **401**, 382-394 (1998). The cultures were incubated at 35°C with a 5% CO_2-enriched atmosphere and fed every other day until use.

After 10-14 days *in vitro*, organotypic cultures were incubated with growth medium containing either 20 μM N-CBZ-L-phenylalanyl-L-alanine-diazomethylketone (ZPAD; BACHEM Bioscience, Torrance, CA), a selective inhibitor of cathepsins B and L (Shaw and Dean, 1980), in 0.01% DMSO, 20 μM chloroquine (Sigma) or vehicle alone for days as specified. To test the effect of EA-1 on the generation of hyperphosphorylated tau fragments found in neurofibrillary tangles in Alzheimer's disease and other tau pathology-related diseases, 1 μM of EA-1 or 10 μM of CEL5-172 were applied alone or together with 20 μM ZPAD.

2. Immunoblotting

For western blot, entorhinohippocampal explants were collected and sonicated in 10 mM Tris-HCl buffer (pH 7.4) containing 0.32 M sucrose, 2 mM EDTA,